peqlab

Cellometer® Auto 1000

User Manual

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Cellometer Auto 1000 User Manual

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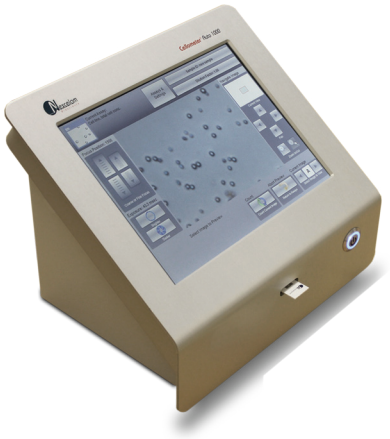
Introduction

What is Cellometer Auto 1000?

Cellometer® Auto 1000 is a compact, automated cell counting system to count, measure cell size and calculate cell concentration from a 20µL cell sample. The basic principle of the Cellometer automatic cell counter is imaging cytometry. Cells are loaded into the Disposable Counting Chamber and automatically spread into a thin layer by capillary action. Cellometer Auto 1000 then captures images of cells in the counting chamber, analyzes the number of cells and sizes of each cell, and then converts this data into concentration, size and viability.

The Cellometer Auto 1000 system consists of 2 main components:

1. Cellometer Auto 1000 instrument with analyzing software



2. Disposable Counting Chambers

- Disposable counting chamber accommodates 2 individual samples and can be loaded through either port.
- Pipette 20µL of cell sample into one of the ports with any standard single channel pipette.
- Cellometer Auto 1000 comes with a starter set of 75 slides. Slides can be ordered directly from Nexcelom or your authorized Nexcelom dealer.

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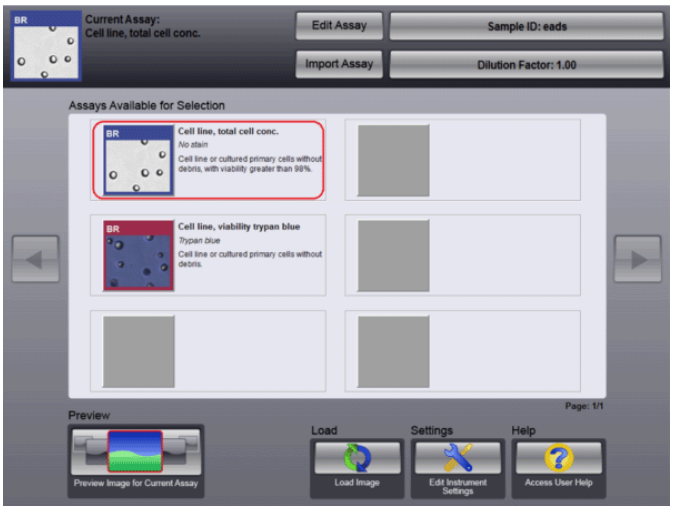
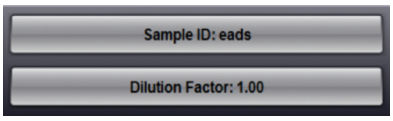
Quick Operation Instructions

1. Select an Assay

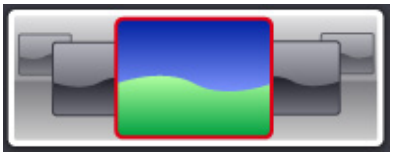


2. Input Sample ID and Dilution Factor

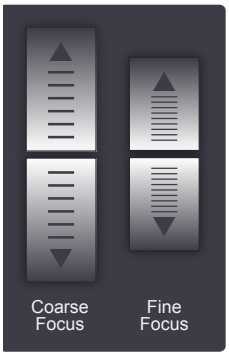


3. Prepare Sample, load disposable counting chamber and insert into instrument

4. Click "Preview"



5. Adjust Focus



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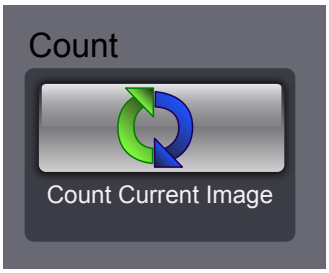
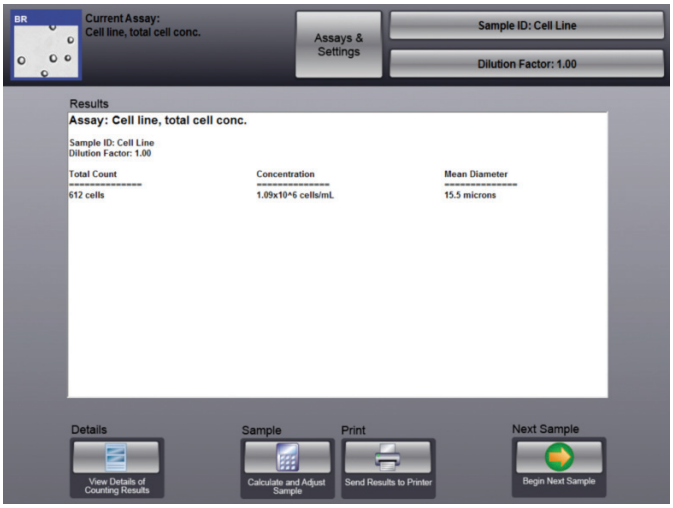
6. Select image to preview and adjust exposure if needed

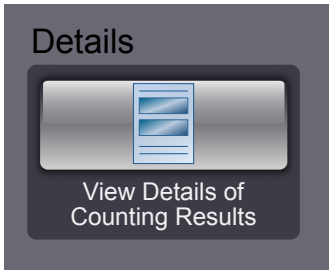
7. Click "Count" to begin counting process



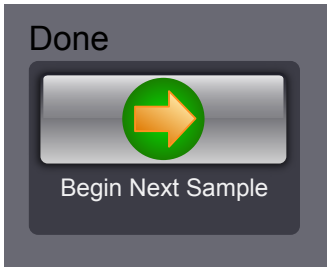
8 .Review Counting Results



9. Select Details to review cell images and counted cell images



10. Select Next Sample or Assay and Settings when done



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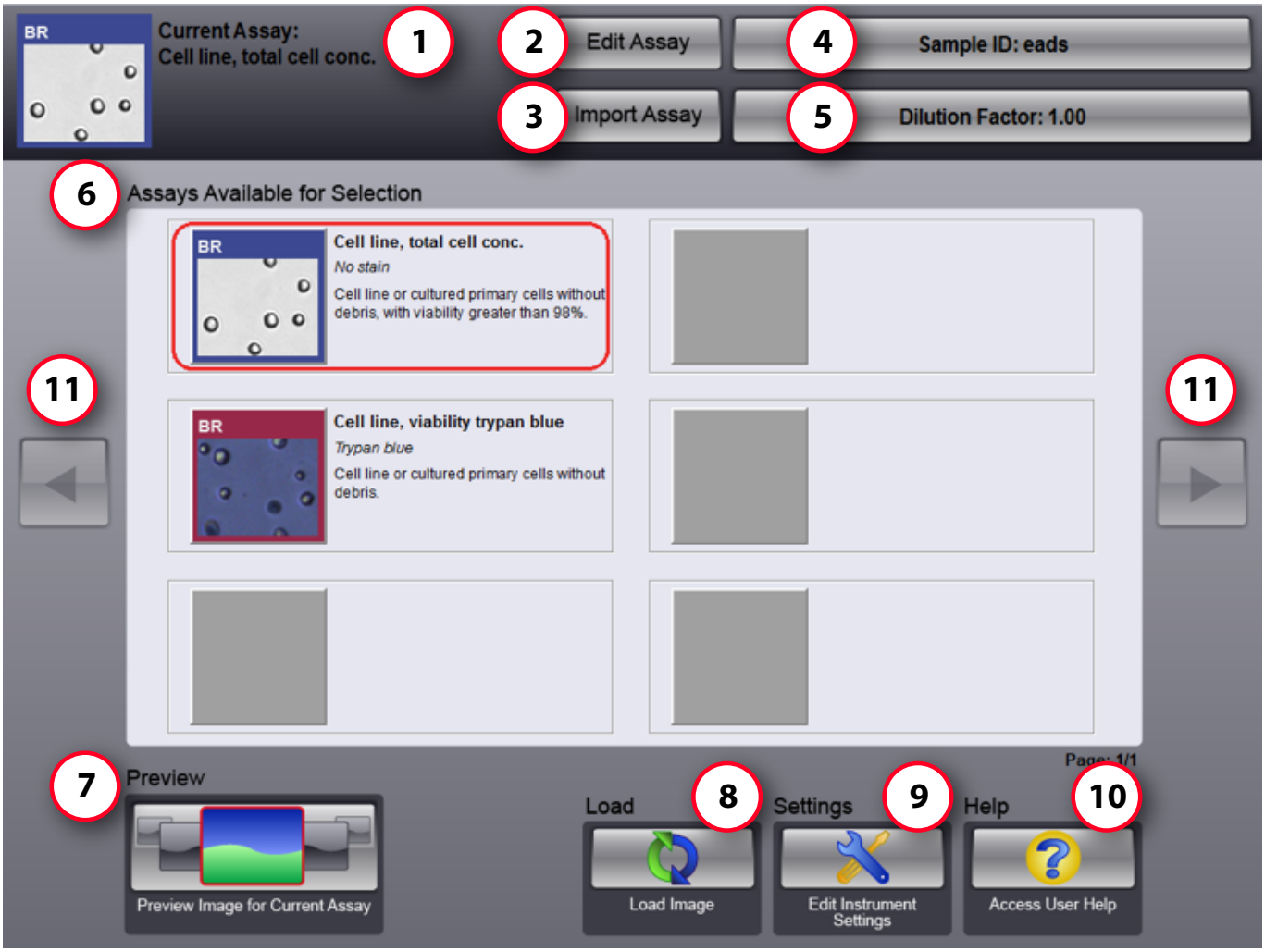
User Interface - Assay Selection Screen



1. **Current Assay** Indicates which Assay is currently selected to analyze a cell sample
2. **Edit Assay** Select to edit the Assay and Cell Type settings
3. **Import Assay** Select to Import additional assays
4. **Sample ID** Select to enter a Sample ID
5. **Dilution Factor** Select to enter a Dilution Factor
6. **Assays Available** List of Assays available for selection
7. **Preview** Select to Preview the cell sample image
8. **Load** Select to load and analyze previously saved images
9. **Settings** Select to edit instrument and UI settings
10. **Help** Select to access help options
11. **Left/Right Arrows** Select to page left/right to access all assays available

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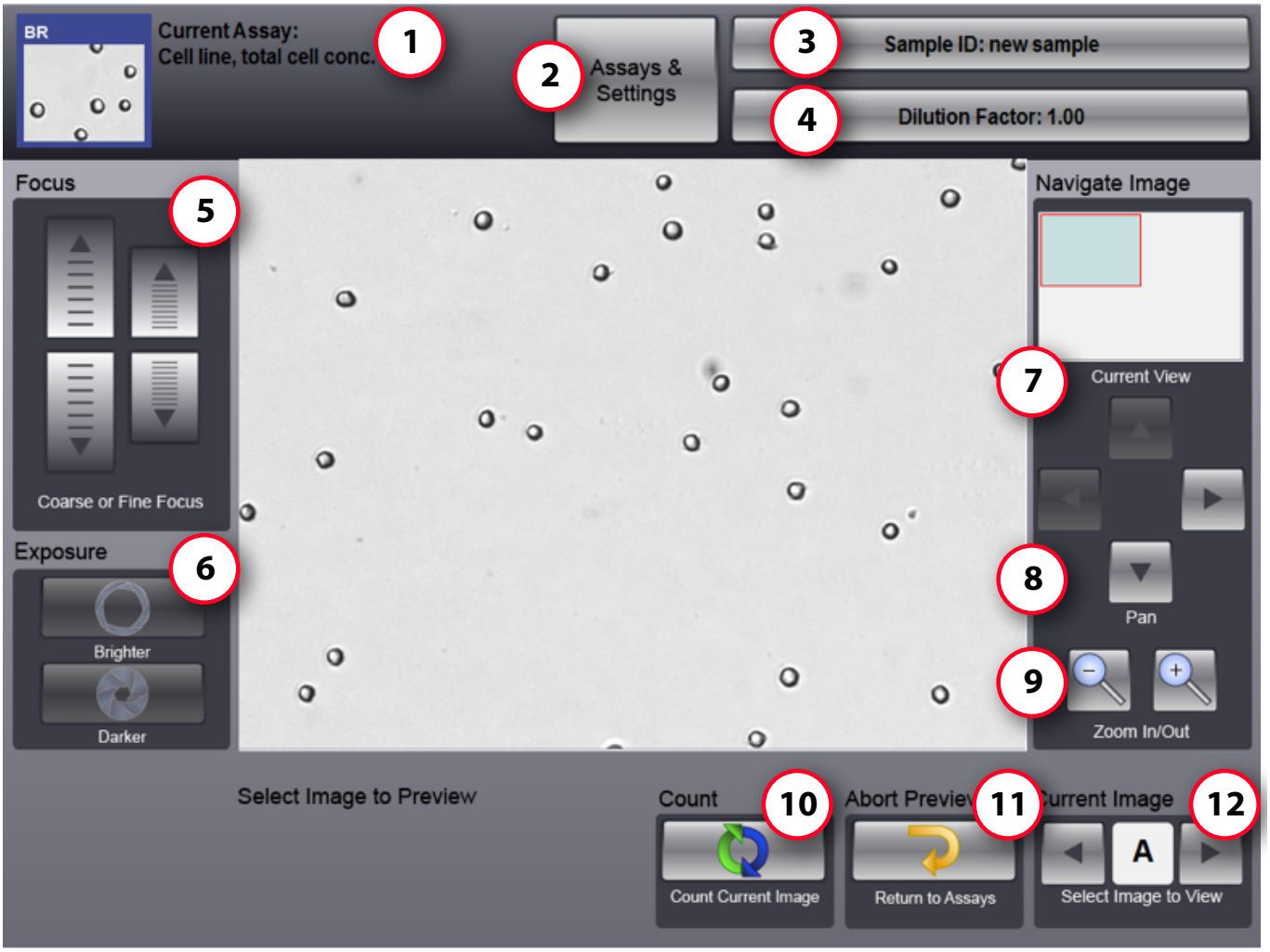
Preview Screen



1. **Current Assay** Indicates which Assay is currently selected to analyze a cell sample
2. **Assays & Settings** Select to return to Assay Selection Screen
3. **Sample ID** Select to enter a Sample ID
4. **Dilution Factor** Select to enter a Dilution Factor
5. **Focus** Fine and coarse focus adjustment of the current cell image
6. **Exposure** Adjust the exposure time of the current cell image
7. **Current View** Red rectangle to show what area of the cell image is currently displayed
8. **Pan** Moves the area currently being shown within the current cell image
9. **Zoom** Zooms in or out of the current cell image
10. **Count** Starts counting of the current cell image
11. **Abort Preview** Cancels current cell preview and returns Assay Selection Screen
12. **Current Image** Select location on slide to preview

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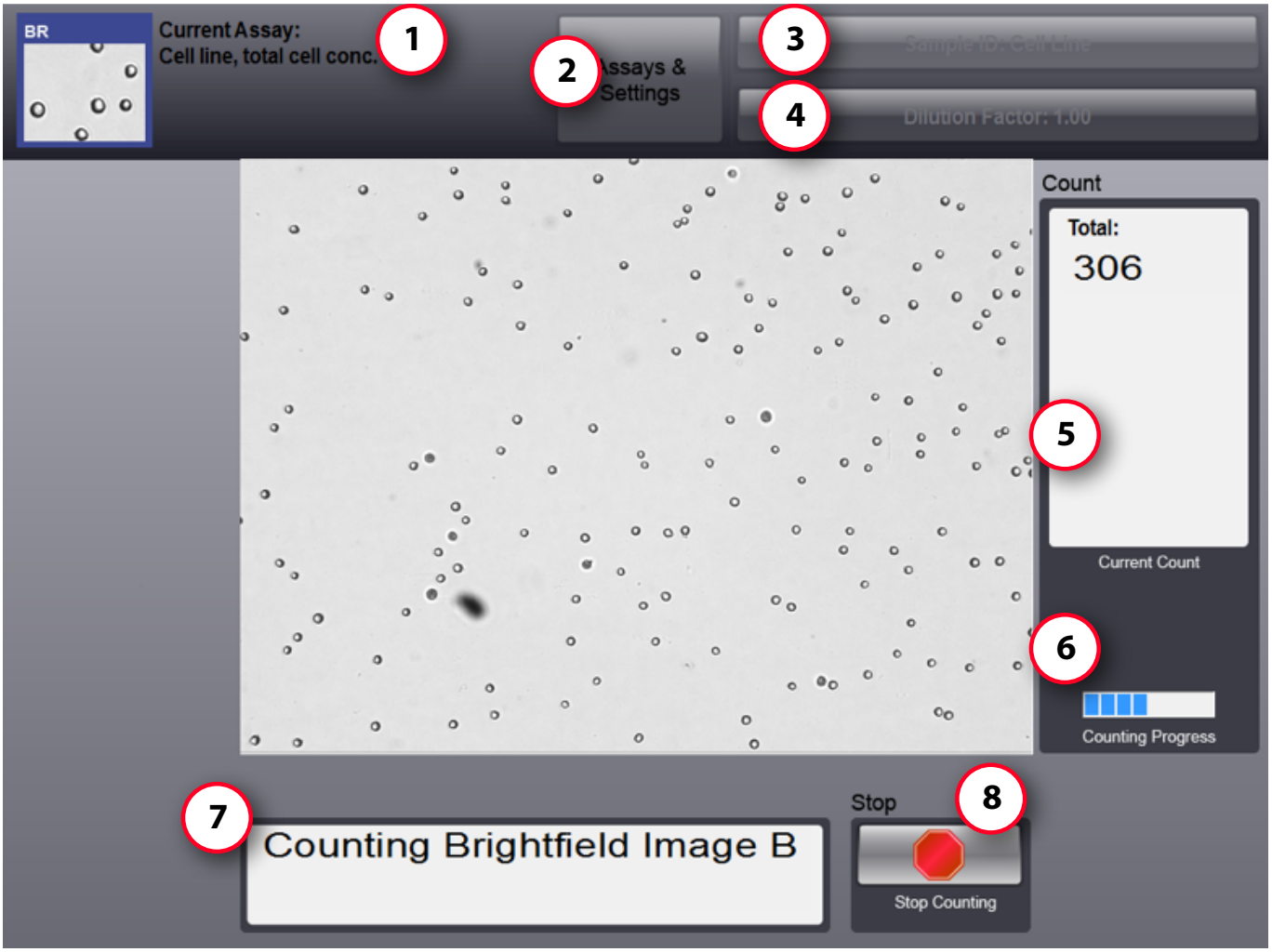
Counting Screen



1. **Current Assay** Indicates which Assay is currently selected to analyze a cell sample
2. **Assays & Settings** Select to return to Assay Selection Screen (disabled while count is in progress)
3. **Sample ID** Select to enter a Sample ID (disabled while count is in progress)
4. **Dilution Factor** Select to enter a Dilution Factor (disabled while count is in progress)
5. **Current Count** Shows cell count numbers
6. **Counting Progress** Visually indicates the current count progress
7. **Image Being Counted** Indicates which image is currently being counted
8. **Stop** Stops cell counting

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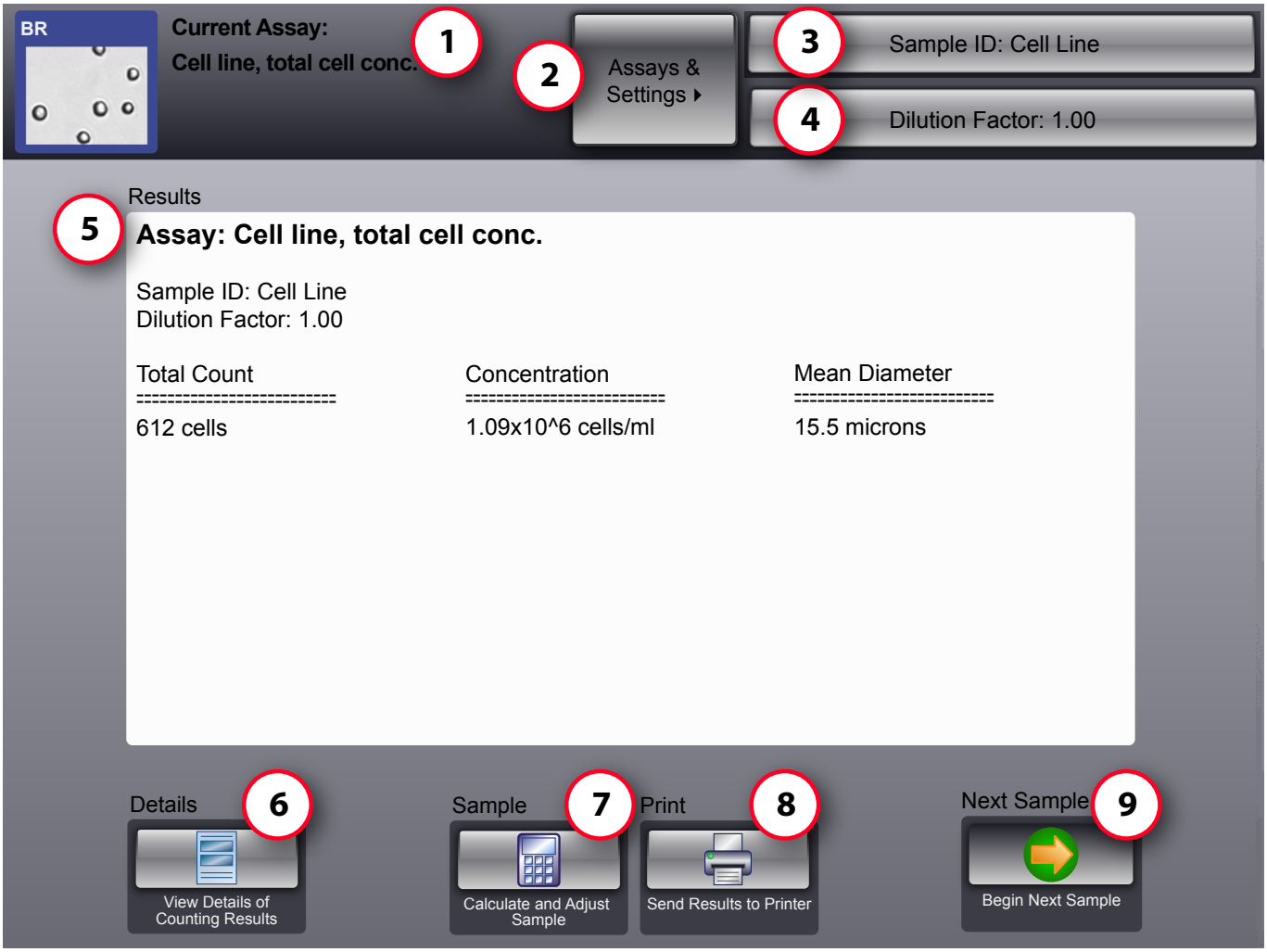
Results Screen



1. **Current Assay** Indicates which Assay is currently selected to analyze a cell sample
2. **Assays & Settings** Select to return to Assay Selection Screen
3. **Sample ID** Select to enter a Sample ID
4. **Dilution Factor** Select to enter a Dilution Factor
5. **Results** Displays cell counting results including total cell count, live/dead cell count and viability
6. **Details** Select to go to the Details Screen
7. **Sample Adjustment Calculator** Select to launch the sample adjustment calculator. Useful for sample adjustment to get desired concentration or total cell number.
8. **Print** Send the counting results to a network printer
9. **Done** Select to start a preview of a new cell sample using the same Assay settings

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Calculate and Adjust Sample Screen

1. **Sample Adjust Source** Select which concentration source to use for the Sample Adjustment calculations
2. **Original Sample Volume** Select to indicate the original sample volume of the cell sample
3. **Target** Select to indicate the target concentration or number of cells which should be calculated

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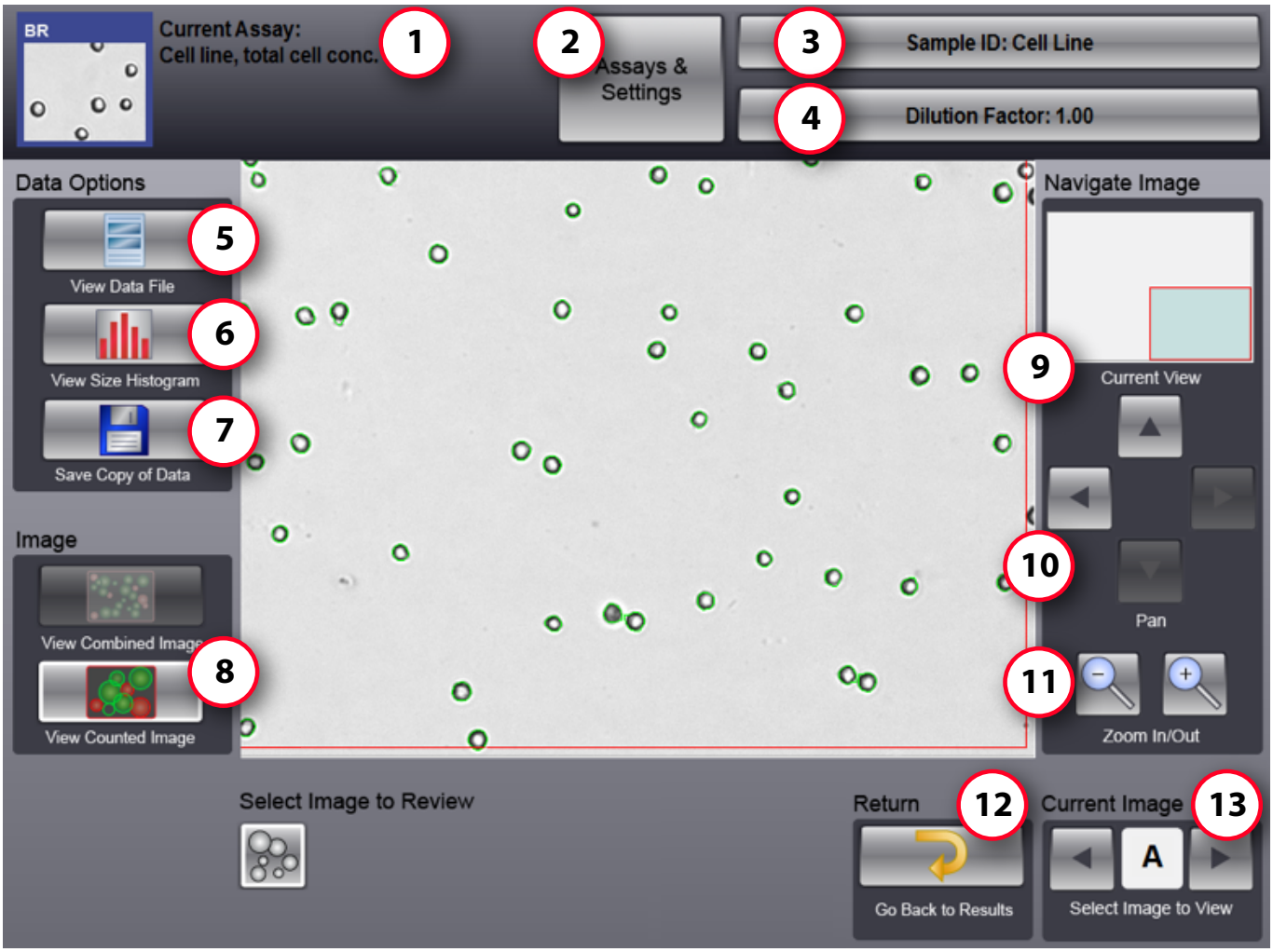
Details Screen



1. **Current Assay** Indicates which Assay is currently selected to analyze a cell sample
2. **Assays & Settings** Select to return to Assay Selection Screen
3. **Sample ID** Select to enter a Sample ID
4. **Dilution Factor** Select to enter a Dilution Factor
5. **View Data File** Opens data file to view or print
6. **View Size Histogram** Opens separate window to display cell size histogram
7. **Save Copy of Data** Select to saves the current counting results and images
8. **View Counted Image** Shows cell image with green and red outlines to indicate cells counted
9. **Current View** Red rectangle indicates what area of the cell image is currently displayed
10. **Pan** Moves the area currently being shown within the current cell image
11. **Zoom** Zooms in or out of the current cell image
12. **Return** Select to return to Results Screen
13. **Current Image** Select location on slide to view counted image

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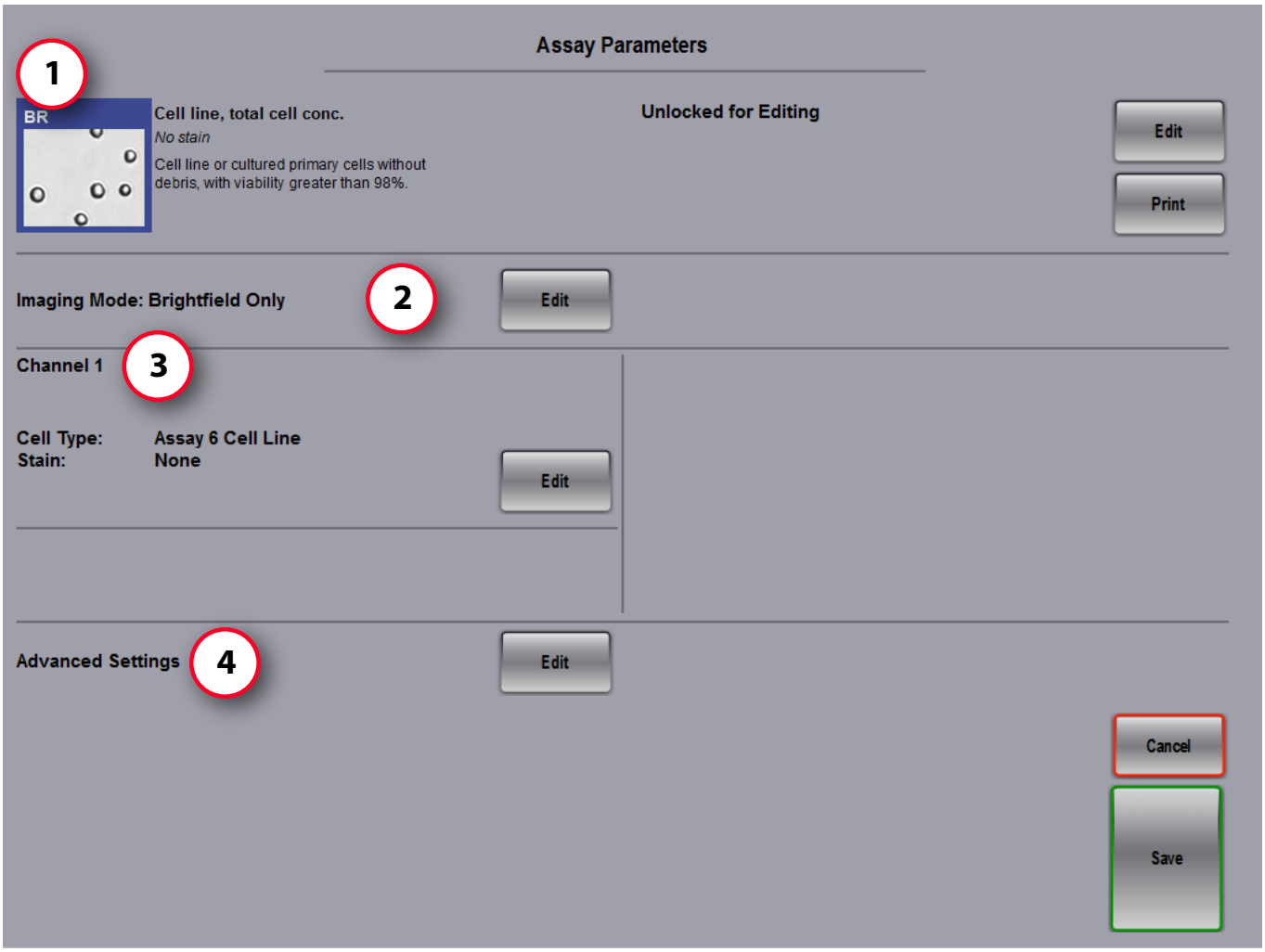
Assay Editor Main Screen



1. **Current Assay** Name of assay, description and icon that appears in the Assay Selection screen
2. **Imaging Mode** Select to indicate which type of images are captured and analyzed
3. **Channel 1** Indicates which Cell Type parameters are used based on the Imaging mode selected.
4. **Advance Settings** Select to edit additional settings available for the imaging mode selected.

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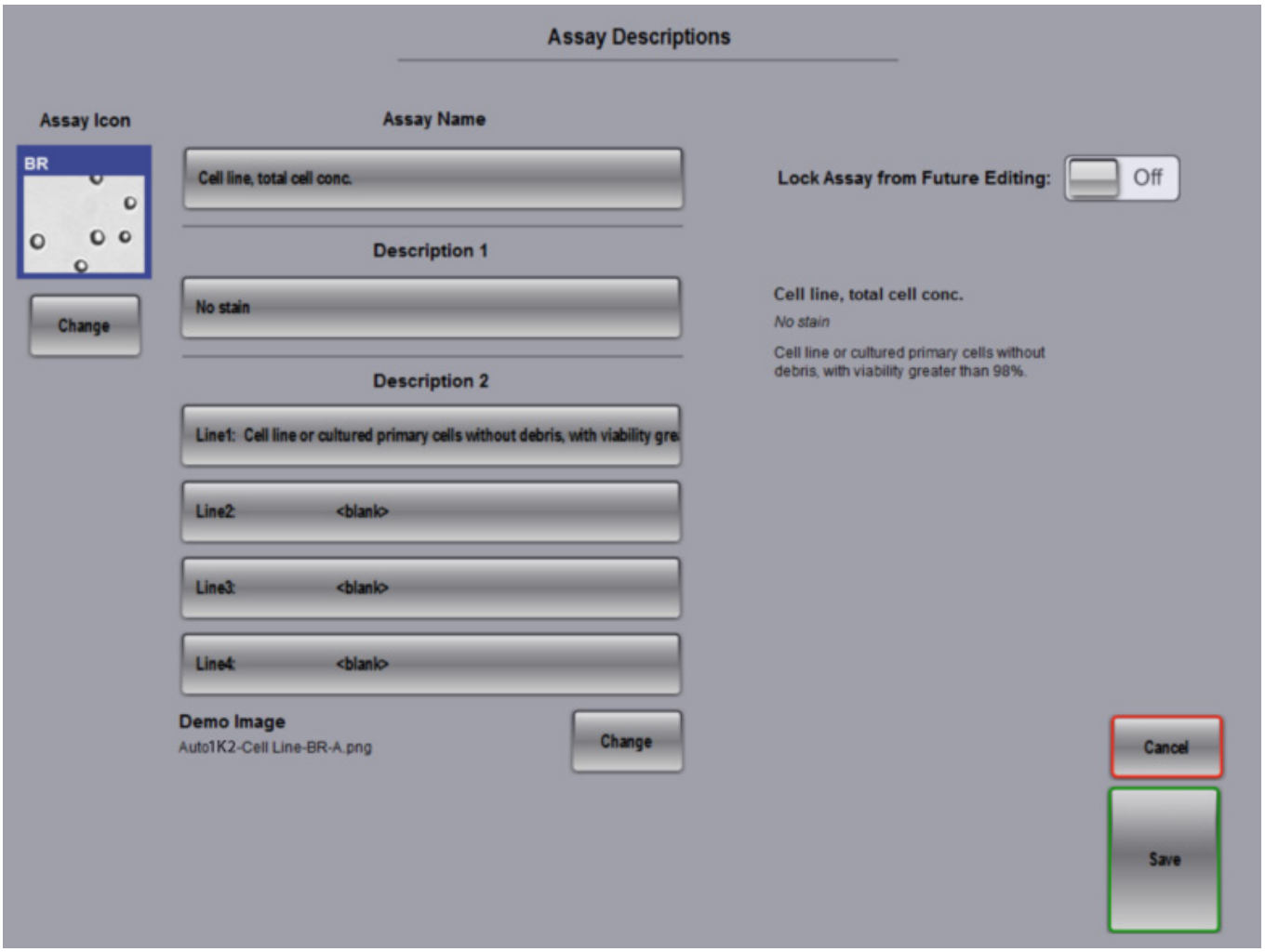
Assay Description Screen



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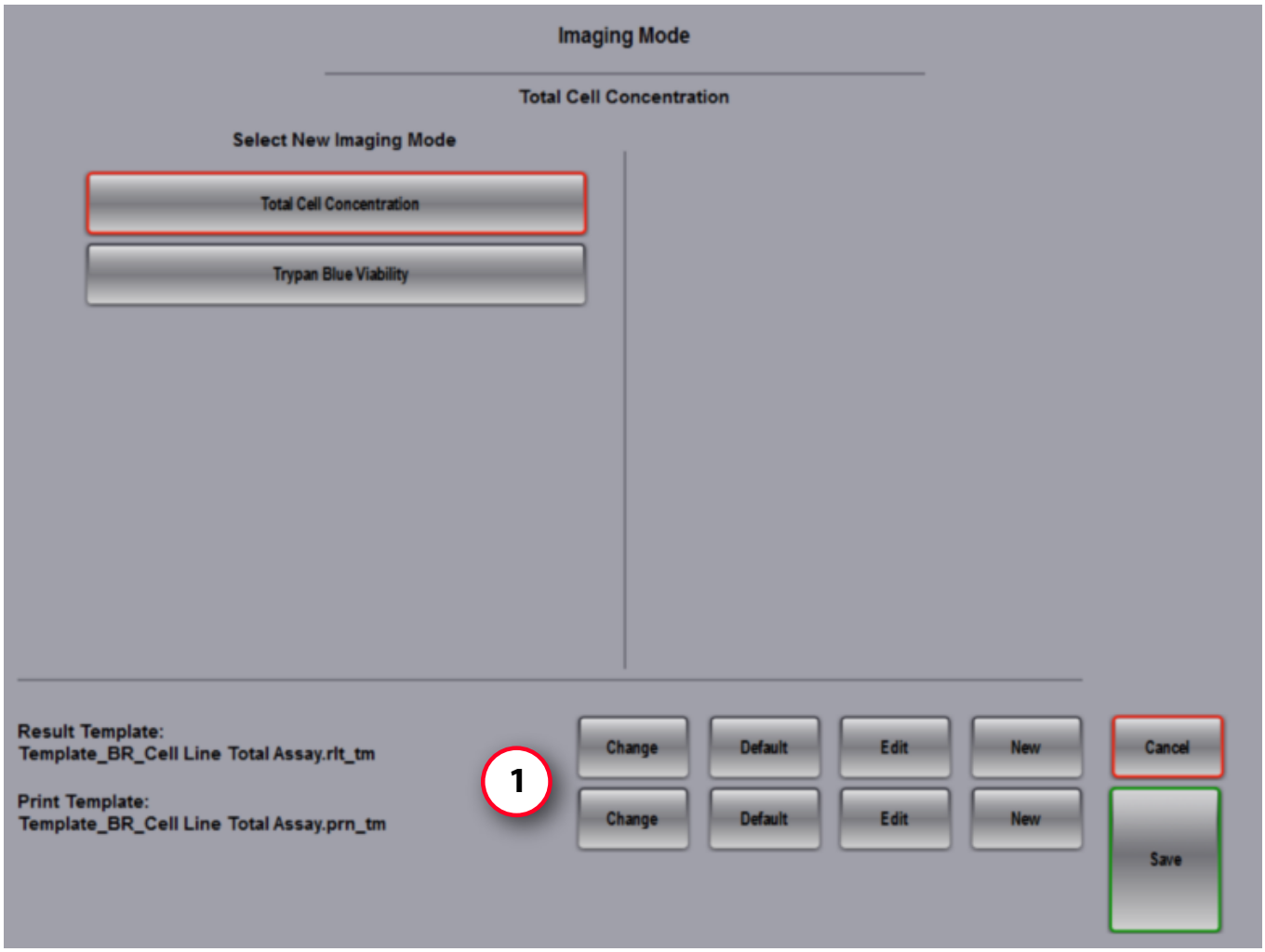
Imaging Mode Screen



1. **Calculations and Reports** Select to edit the report formats and the calculations used to generate results.

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Channel Settings Screen

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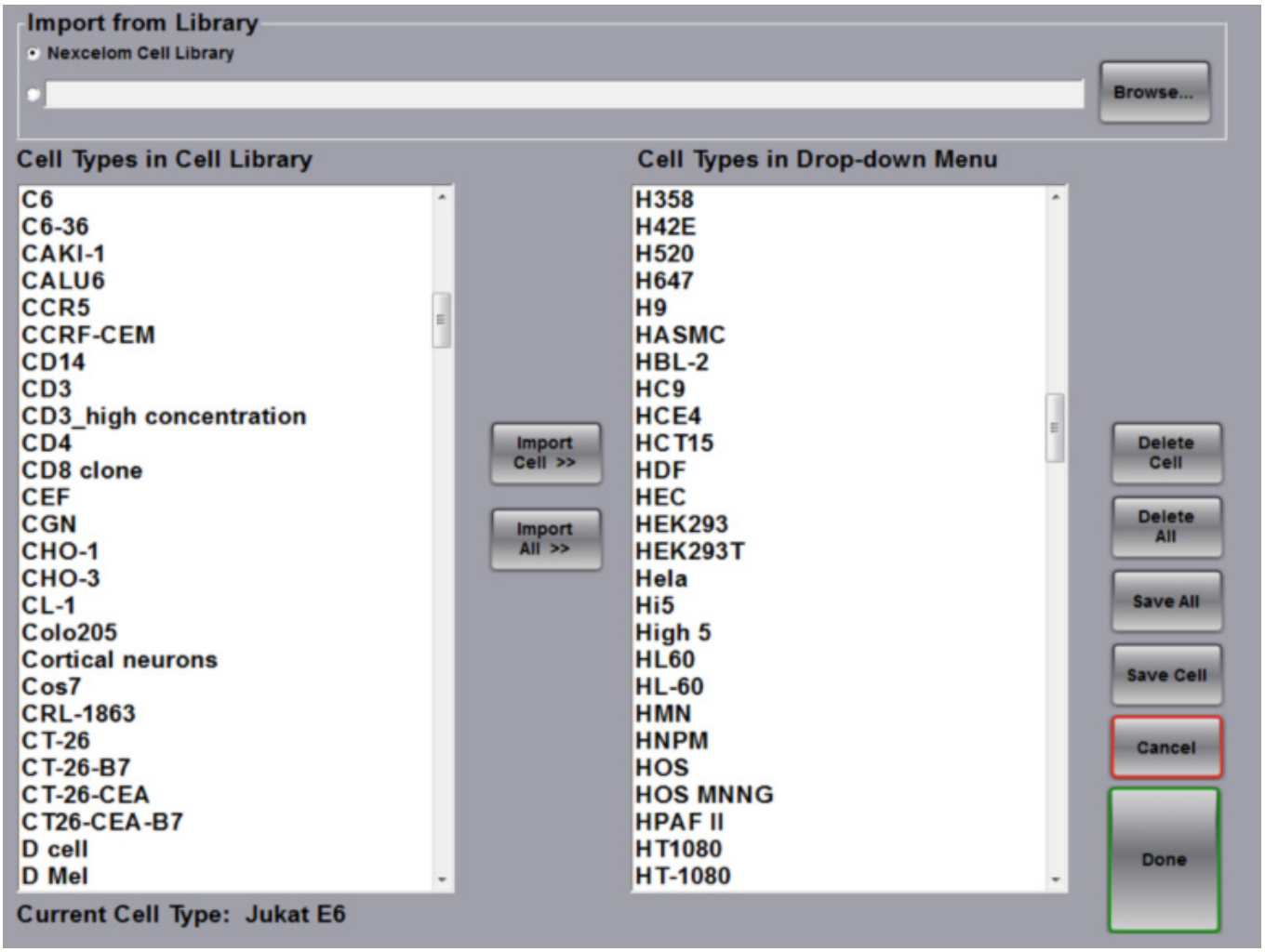
Cell Type Library Screen



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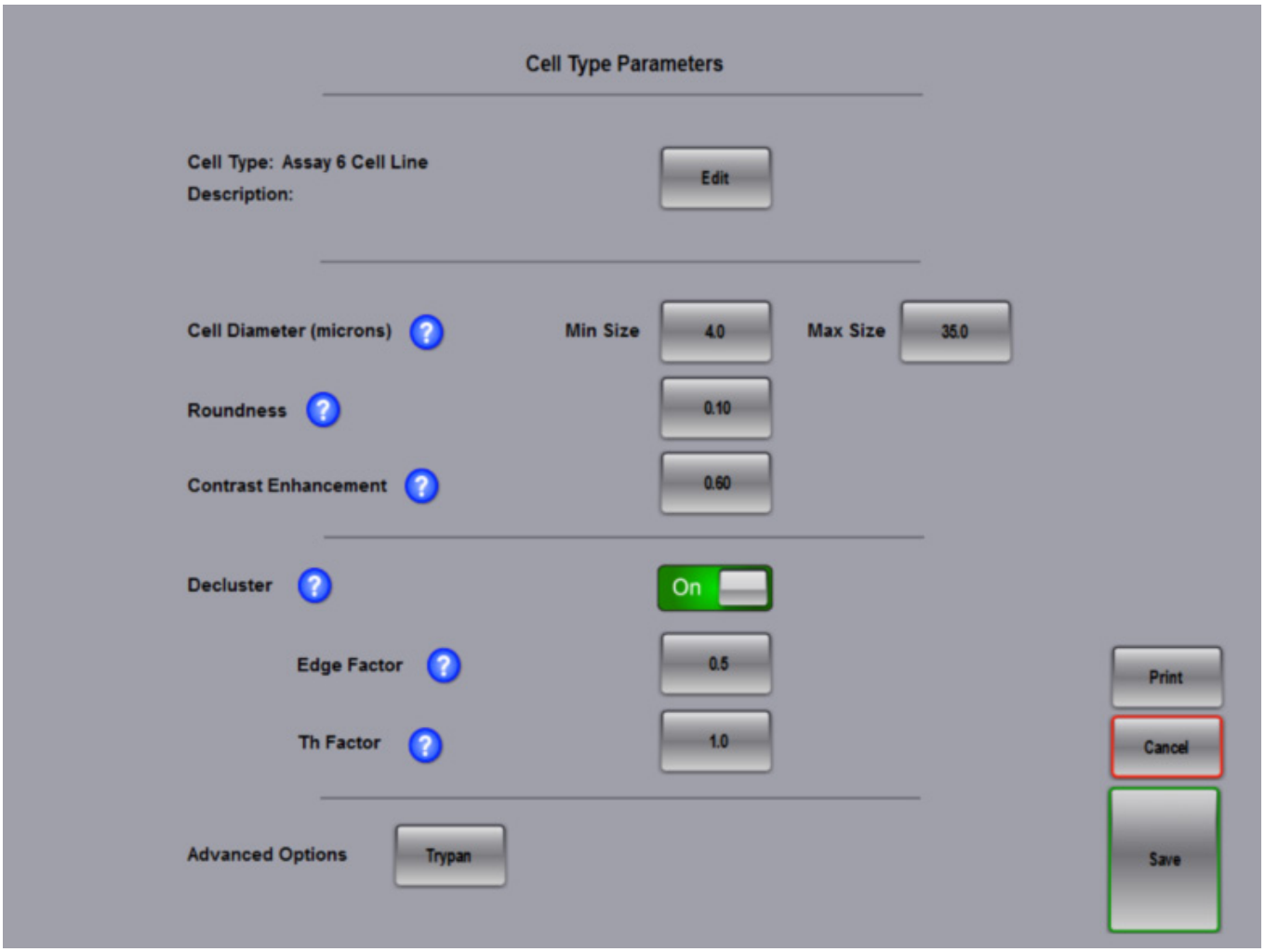
Cell Type Parameters Screen



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Getting Started

All Nexcelom products undergo a rigorous quality inspection prior to shipment and all reasonable precautions are taken in preparing them for shipment to assure safe delivery.

The instrument should be unpacked and inspected for mechanical damage upon receipt. Mechanical inspection involves checking for signs of physical damage such as scratched, dents, etc.

If damage is apparent, or any components are missing, please immediately contact Nexcelom (+1-978-327-5340 or support@nexcelom.com) or your local dealer.

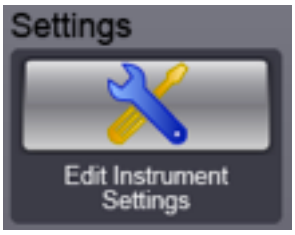
After unpacking the instrument, plug the Cellometer Power Cable into the back of the instrument. The Auto 1000 is pre-configured with the Cellometer Auto 1000 software. No additional setup or configuration is required.

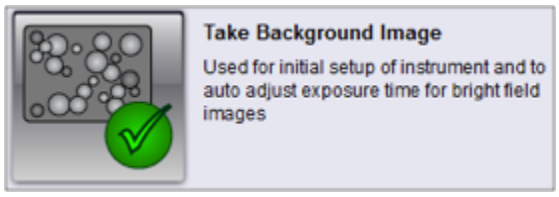
Starting the Auto 1000

After plugging in the system, simply press the power button on the front of the unit.

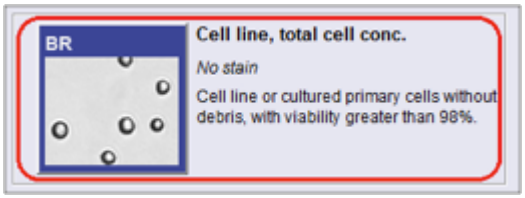
Taking a Background Image

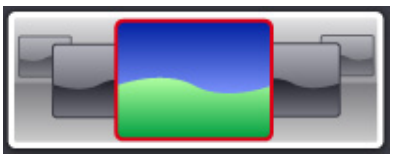
1. When using the Auto 1000 for the first time, a background image should be taken. This step does not need to be repeated unless the instrument is moved.
2. Confirm that there is no slide in the Auto 1000 instrument.
3. Click on the "Settings" button, then take the "Take Background Image" button





4. Check the background image by clicking on the Cell Line, Total Cell Conc. assay and the Preview Image button. The display should show an even white screen. If any dark areas or variations are present, contact Nexcelom Technical Support.





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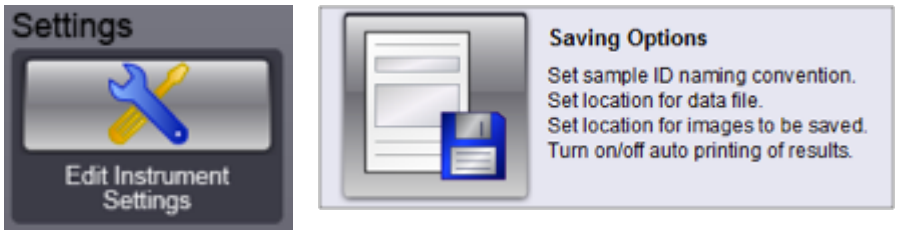
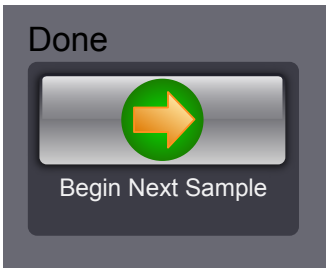
Auto-Save Set-up

1. Insert a USB drive or connect the Auto 1000 to a network drive via an Ethernet network cable.
2. Click on the "Settings" button, then the "Saving Options" button.


Settings
Edit Instrument Settings
Saving Options
Set sample ID naming convention.
Set location for data file.
Set location for images to be saved.
Turn on/off auto printing of results.

3. Click "Auto Save to Data File" and "Save Raw Images" to the "On" position. Click on the File and Folder buttons to set the locations for saving. In each case, select the USB drive or network drive and name the folder. Having saved images will help if any remote technical support is required and provides a permanent record of the raw data. Click "Done", then click "Done" again.

4. Click "Done", then click "Done" again.



Preparing Reference Beads

1. Though the Auto 1000 instrument does not require any routine testing or calibration, reference beads are available.
2. Cellometer Check Validation Bead Solution, product number CCBM-011-2ML, is recommended for use with the Auto 1000.
3. Invert the reference bead solution a total of ten times
4. Vortex the bead solution for ten seconds
5. Set pipette to 20 µl
6. Pipette bead solution up and down ten times to break up any bead clumps

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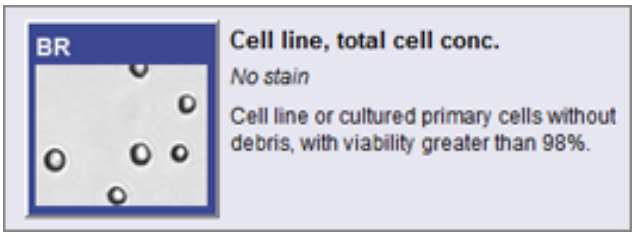
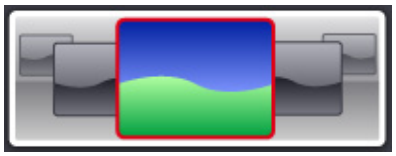
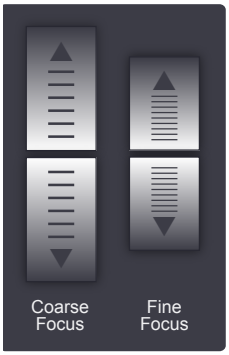
Counting Reference Beads

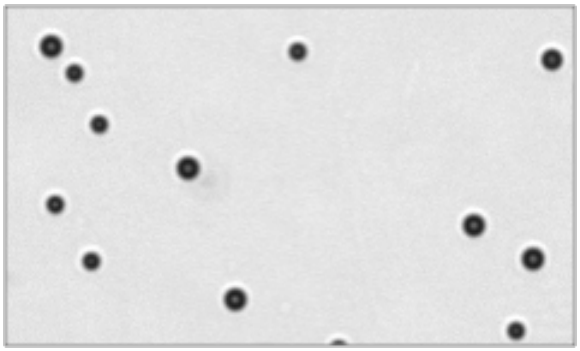
1. Peel plastic off of both sides of the Cellometer slide. (for PD100 slides, the plastic has already been removed)
2. Place cell counting chamber on a fresh Kimwipe
3. Load 20 µl of mixed Reference Bead Solution into the Cellometer Counting Chamber.
4. Insert the loaded chamber into the Auto 1000 sample slot and gently push the slide to the stop.
5. Select the Cell line, total cell conc. assay.



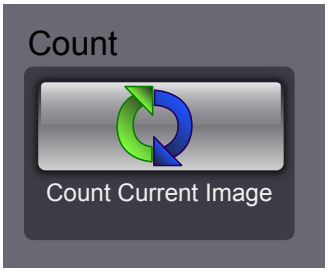


6. Click on "Preview Image for Current Assay"
7. Adjust the focus if necessary using the coarse and fine adjustments on the left-hand side of the screen until the best bead counting focus is achieved. The beads should appear as dark circles with sharp edges.





8. Click the **Count** button at the bottom of the screen.



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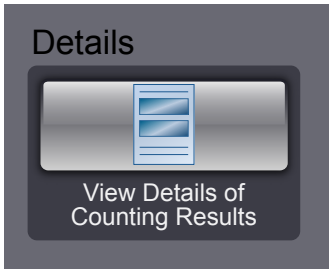
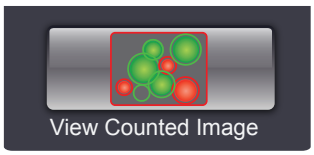
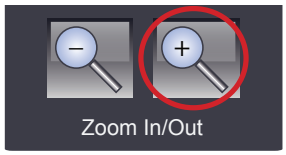
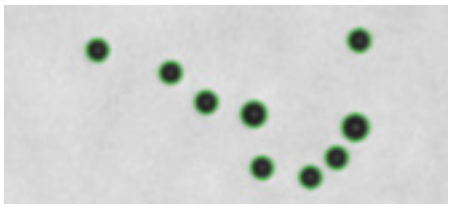
9. When counting is complete, click on the "View Details" button at the bottom left of the screen.



10. Click on the "View Counted Image" button on the left-hand side of the screen. Enlarge the image by clicking the "Zoom In" button on the right-hand side of the screen. Confirm that all of the beads are circled in green.







Comparing Viability Methods

Comparing Methods: When evaluating different viability methods, it is critically important to use one aliquot from the stock cell culture to perform all testing. The cell sample should be evaluated for concentration on the Cellometer Auto 1000 prior to staining.

Trypan Blue Viability: For *cell lines and cultured primary cells,* bright field imaging and trypan blue viability may be used to determine the number, concentration, and percentage of live cells. The trypan blue method is *not recommended* for samples containing debris, platelets, or red blood cells. Fluorescence is required to accurately differentiate nucleated cells from platelets, red blood cells, and debris.

Preparing Cell Sample for Trypan Blue Viability Determination

1. A cell concentration of 1.0 x 10^5 to 1.0 x 10^7 cells/mL can be analyzed on the Auto 1000. A concentration of 1.0 x 10^6 cells/mL is optimal.
2. Invert the tube containing cells ten times and pipette up and down ten times to generate a homogeneous cell sample and reduce cell clumps. Do not shake or vortex the sample! This will generate bubbles.
3. For viability measurement, stain cells by combining 50 µl of cell sample with 50 µl of a 0.2% trypan blue staining solution (for a final concentration of **0.1% trypan blue**). Gently mix by pipetting up and down ten times.

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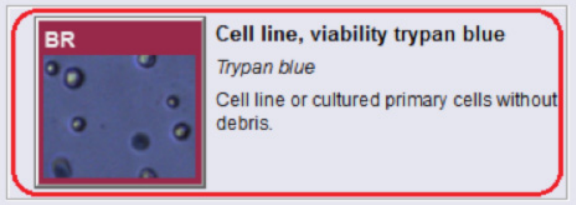
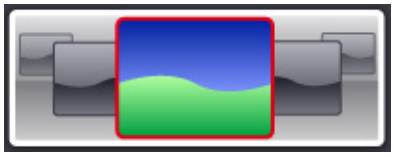
Cell Concentration and Trypan Blue Viability using the Auto 1000

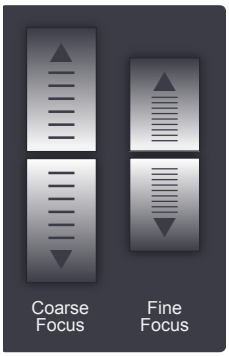
1. Pipette up and down gently 10x to break any potential cell clumps in the stained cell sample
2. Load 20µl of sample into the Cellometer Counting Chamber (CHT4)
3. Insert counting chamber into instrument.
4. Click on the "Cell line, viability trypan blue" assay. When prompted, enter your desired Sample ID and a dilution factor of 2.

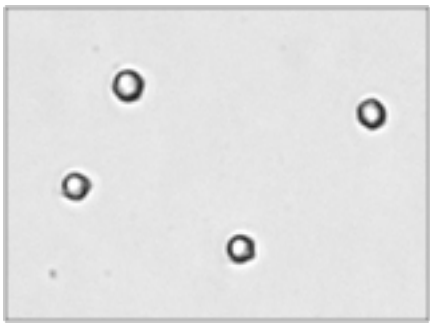


5. Click on "Preview Image for Current Assay"

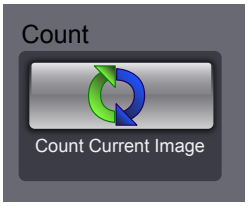


6. Adjust the focus if necessary using the coarse and fine adjustments on the left-hand side of the screen. Cells in focus will have a bright center and crisp edge.

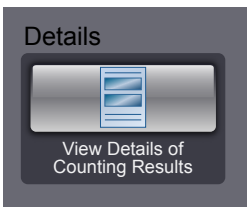




7. Click the Count button at the bottom of the screen.



8. Click on the "View Details" button at the bottom left of the screen, then click on the "View Counted Image" button on the left-hand side of the screen.



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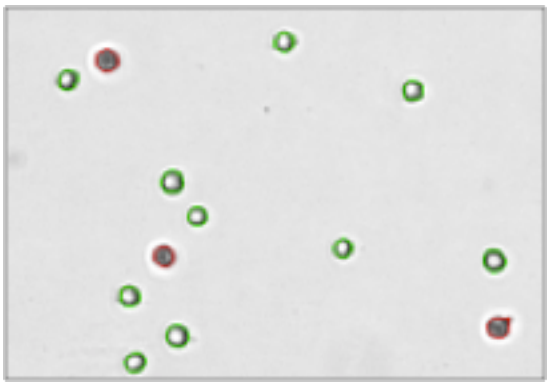
9. Review the counted image. Live cells should be circled in green. Dead cells, stained dark with trypan blue, should be circled in red.



10. The Auto 1000 is now ready to analyze the next sample. After inserting the imaging chamber loaded with the next sample, click on the "Next Sample" button at the bottom right of the screen. When prompted, enter the Sample ID, then click "Count".

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Tutorials

Overview

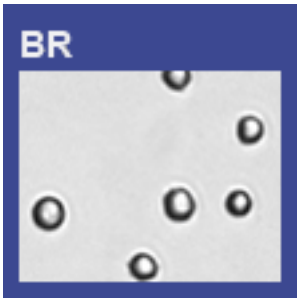
The following tutorials are intended as a guide to performing various cell counting assays using the Auto 1000. General sample preparation hints are included for each tutorial as well as instrument and software operation instructions.

Each of the assays can also be performed using the sample images included in the software as a demonstration of the Auto 1000. Sample images for each cell counting assay can be found at C:\Program Files\Nexcelom\Assay_Images\.

Staining Solutions

Trypan Blue Stock solution: 0.2% in PBS. Use 1:1 with cell sample. Dilution factor: **2**

Installed Assays and Descriptions

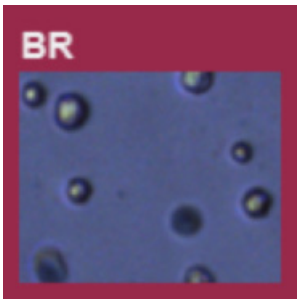


Cell Line, Total Cell Concentration

No Stain

Cell line or primary cells without debris, with viability greater than 98%.

After taking brightfield images of a cell sample, all cells are counted to determine total cell concentration.



Cell Line, Viability Trypan Blue

Trypan Blue

Cell line or cultured primary cells without debris.

Trypan blue is routinely used to determine cell viability. Trypan blue penetrates and stains dead cells and leaves live cells unstained. After taking brightfield images of the stained sample, live and dead cells are counted to determine total, live and dead cell concentrations as well as compute percent viability.

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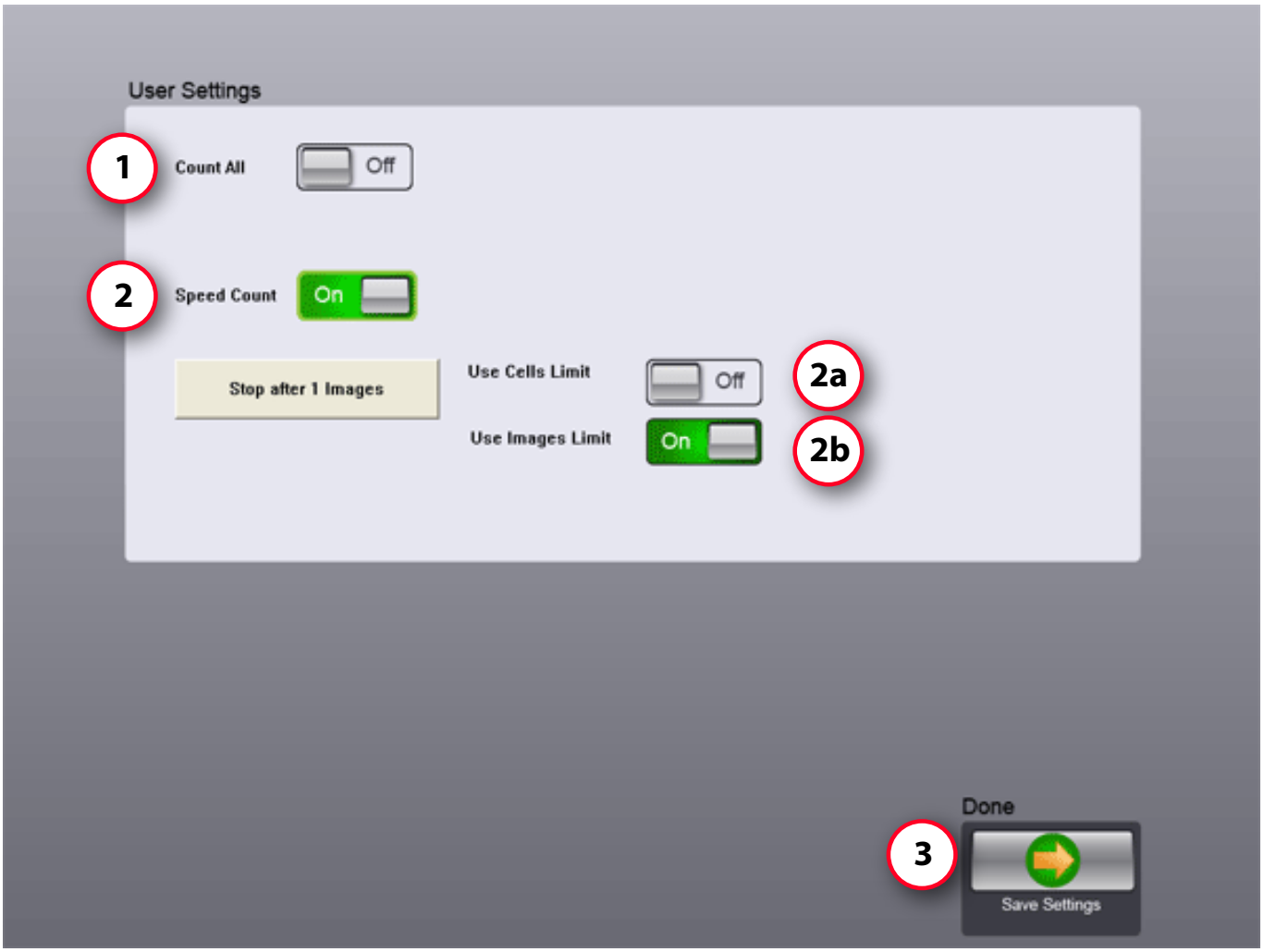
Operation Reference

Counting Options



1. **Count All** Select to have all 4 positions of slide used for cell images
2. **Speed Count** Select to have counting use Cell Limit or Image Limit
   a. **Use Cells Limit** Check to stop counting after # of user defined cells are counted and the next frame has finished counting
   b. **Use Images Limit** Stop counting after finishing user defined images that are less than 4 images
3. **Done: Save Settings** Saves the current settings and returns to previous screen.

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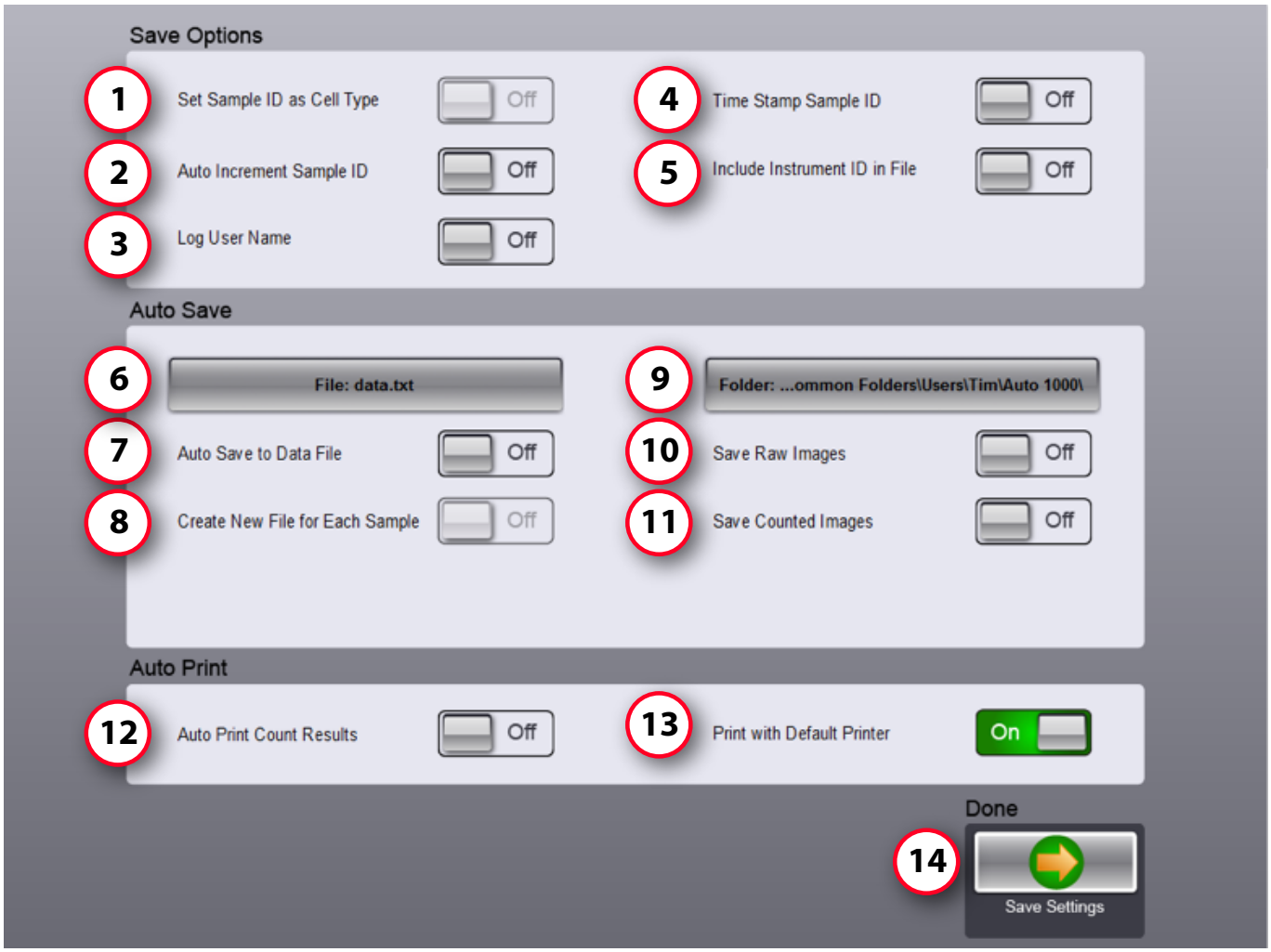
Saving Options



Save Options

1. **Set Sample ID as Cell Type** Selecting this will auto input the Sample ID to match the Cell Type parameter name being used for counting
2. **Auto Increment Sample ID** Selecting this will auto append the Sample ID with an incremental numerical value (Example: CHO sample_001)
3. **Log User Name** Selecting this will require the user to enter in an ID that will be recorded with the data
4. **Time Stamp Sample ID** Selecting this will auto append the Sample ID with the date and time the count was performed
5. **Include Instrument ID in File** Selecting this will auto append the Instrument ID to the sample ID after the count is performed

Auto Save

6. **File** this button brings up the File Dialog Window which allows the user to specify where the data.txt file will be saved when Auto Save to Data File is selected.
7. **Auto Save to Data File** Selecting this will auto save the data into the Data.txt file after a count is performed

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8. **Create New File for Each Sample** Selecting this will auto create a new data.txt file for each sample and save the counting results to it after a count is performed
9. **Folder** this button brings up the File Dialog Window which allows the user to specify where the image folder will be saved when Save Raw Images or Save Counted Images is selected.
10. **Save Raw Images** Selecting this will auto save the images to the folder specified using the Folder command
11. **Save Counted Images** Selecting this will auto save the counted images to the folder specified using the Folder command

Auto Print

12. **Auto Print Results** Select to have the results sent to the printer upon completion of count.
13. **Print with Default Printer** Select to have the Windows default selected printer used when Auto Print Results is selected.
14. **Done** Save Settings Click to save new settings and return to Assay and Settings main screen.

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Technical Information

Specifications

Size and Weight	Height 9.0 inches (229 mm) Width 11.1 inches (282 mm) Depth 8.3 inches (211 mm) Weight 9.0 Pounds (4.1 kg)
Environmental Requirements	Typical biology lab environment
Display	10.4 inch, 1024 x 768 TFT with LED back light
Touch Screen	4-Pin resistive with controller 10.4 inch format
Processor	Intel® Core i3
Memory	RAM Memory: 4 GB DDR2 Hard drive size: 16 or 32 GB Mini PC13 BIOS: 2 MB flash
Operating System	Windows embedded system
External Connectors	2 USB 2.0 1 Ethernet 1 Power input plug
Power Input	12 VDC @ 7.5 Amps
External Button	On/off 2-color button Orange: Standby Blue: Run
Circulation and Cooling	Allow at least 2 inches of unobstructed air circulation around the vent holes on the Auto 1000 while in use to ensure adequate cooling of the product.

Getting Support

We provide free consulting for your lab on cell type, assay type and sample conditions. The goal is to find the most suitable cell counting / analysis solution for your lab.

Our team of Application Specialists are trained biologists with comprehensive understandings of cell counting, viability assay methods and best practices.

Please contact us when you need to discuss specific types of cells, sample conditions, and applications. We are available between 8:30am and 5:00pm Eastern US time. For immediate help, please call us at 1-978-327-5340 or email

support@nexcelom.com for applications and other technical information
sales@nexcelom.com for placing a purchase order or inquiries on a purchase order
info@nexcelom.com for general inquiries

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Warranty Information

Nexcelom warrants that Nexcelom instrumentation products shall, for a period of 12 (twelve) months from the date of purchase, be free of any defect in material and workmanship. The sole obligation of this warranty shall be to either repair or replace at our expense the product, at manufacturers option. The original sales receipt must be supplied for warranty repair. Products, which have been subjected to abuse, misuse, vandalism, accident, alteration, neglect, unauthorized repair or improper installation, will not be covered by warranty.

Any Product being returned is to be properly disinfected and packaged (in original packing if possible). Damage sustained in shipping due to improper packing will not be covered by warranty. A valid Return Material Authorization Number (RMA#) is required for all warranty repairs. For RMA instructions, please contact our customer service department at 978-327-5340 or email support@nexcelom.com.

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Limitation of Liability (Hardware and Software)
Cellometer® branded automatic cell counting instruments, software and consumables are intended for research use only.

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